ABSTRACT

A method is disclosed for improving encapsidation of transgene RNA using retroviral packaging and transfer vectors. An HIV-2 transfer vector, which includes the transgene, is introduced into a packaging cell that is also transfected with (or stably expresses) an HIV-2 derived packaging vector or a combination of packaging vectors. The packaging vector has mutations in packaging signal sequences that are both upstream and downstream of the 5' splice donor site. The upstream mutation can be a functional deletion of a signal sequence located between the 5' LTR and the 5' splice donor site, while the downstream mutation can be a functional deletion of a signal sequence located between the 5' splice donor site and an initiation codon of the gag gene on the HIV-2 genome. It can also be composed of a combination of two or more partial vectors. A transfer vector, which is introduced into the packaging cell line, has a mutation that renders its splice donor site non-functional. Transgene RNA expression and encapsidation from these cells is markedly increased, but with little or no levels of infectious viral RNA encapsidation. In particular embodiments, the packaging vector is an HIV-2(ROD) clone, such as pROD(SD36) or pROD(SD36/EM) plus pCM-ENV(ROD), the transfer vector is an HIV-2 clone, such as pSGT-5(SDM), and the packaging cell is a 293T cell. The invention also includes vectors used in this method, cells transformed with the vector or vectors, the supernatant of the packaging cells with the encapsidated vector RNA, as well as the encapsidated RNA itself.